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(54) PRODUCTION OF FUNCTIONAL PROTEINACEOUS MATERIAL

(57) Abstract:

PURPOSE: To obtain a method for producing a functional proteinic material soluble in organic solvents.

CONSTITUTION: The objective method for producing a functional proteinic material is characterized by mixing at least two proteins, water and a crosslinking agent, crosslinking the proteins and reacting the resultant crosslinked proteins with at least one selected from an alkylating agent, a Schiff base-forming agent and an acid.

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(54)【発明の名称】　機能性蛋白質素材の製造法

(57)【要約】
【目的】　本発明の目的は、有機溶媒に可溶な機能性蛋白質素材の製造法を提供することにある。
【構成】　本発明の機能性蛋白質素材の製造法は、少なくとも２種の蛋白質、水及び架橋剤を混合、架橋させ、次いで得られる蛋白質の架橋物にアルキル化剤、シッフ化剤及び酸から選ばれた少なくとも１種を反応させることを特徴とするものである。

【特許請求の範囲】
【請求項１】　少なくとも２種の蛋白質、水及び架橋剤を混合、架橋させ、次いで得られる蛋白質の架橋物にアルキル化剤、シッフ化剤及び酸から選ばれた少なくとも１種を反応させることを特徴とする機能性蛋白質素材の製造法。
【請求項２】　蛋白質が遊離アミノ基を有する蛋白質又はペプチドである請求項１記載の機能性蛋白質素材の製造法。
【請求項３】　遊離アミノ基を有する蛋白質又はペプチドが鶏、うずら、あひる又はガチョウの卵の卵白蛋白質、ホエー蛋白質、血清アルブミン及びカゼインから選ばれた少なくとも１種である請求項２記載の機能性蛋白質素材の製造法。
【発明の詳細な説明】
【０００１】
【産業上の利用分野】本発明は、機能性蛋白質素材の製造法に関する。更に詳しくは、本発明は、有機溶媒に可溶な機能性蛋白質素材の製造法に関する。
【０００２】
【従来技術とその課題】従来から合成繊維の肌触り、湿気の吸入性や放出性、保温性等を改良するため、合成繊維に蛋白質を添加する試みがなされている。
【０００３】例えば、特開平１－２９３１４３号公報によれば、ゼラチンと絹の微粉末を合成樹脂に分散させたものが提案されているが、微粉末の粒径にバラツキがあって微粉末が樹脂中で均一に分散しないため、該合成樹脂から製造される合成繊維は肌触り、湿気の吸入性と放出性のバランス、保温性等の点で満足できるものではない。しかもゼラチンと絹の微粉砕化は、粉砕の操作の煩雑さ、粉砕品の飛散する等して、取扱いが大変難しいという問題を有している。
【０００４】而して、樹脂中での蛋白質の分散性を改良するには、樹脂を溶解する有機溶媒に溶解し得る蛋白質があれば、好ましい結果が得られることは明白である。
【０００５】有機溶媒に溶解し得る蛋白質としては、例えば、単純蛋白質のプロラミンが知られているが、これは６０～９０％のエタノール（即ち水性有機溶媒）に可溶であるにすぎず、９０％以上のより純度の高いエタノール及び他の有機溶媒には不溶であるため、実用に供し得ない。
【０００６】また特開昭５２－２５８００号公報には、蛋白質とイソシアネート化合物の付加反応物を有機溶媒又は水性有機溶媒の存在下に加水分解してなる改質蛋白質がジメチルスルホキシド（ＤＭＳＯ）、ジメチルアセトアミド（ＤＭＡ）等の有機溶媒に可溶である旨記されている。しかしながら、斯かる蛋白質は赤外部吸収で１７４０ｃｍ$^{-1}$付近、１２２２ｃｍ$^{-1}$付近のアミド結合やウレタン結合を有する側鎖が切断され尿素結合を有する側鎖のみが残存した、所謂低分子化された蛋白変性物であるため、蛋白質としての機能が低下する虞れがある。しかも、斯かる改質蛋白質の有機溶媒への溶解性も充分とは言えない。
【０００７】
【発明が解決しようとする課題】本発明者は、上記従来技術の現状に鑑み、鋭意研究を重ねた結果、１種の原料蛋白質をジイソシアネート等の特定の架橋剤で架橋高分子化することにより、有機溶媒に可溶な蛋白質素材が得られることを見い出し、先に特許出願した（特願平４－５３４６６号）。
【０００８】上記出願に係る発明により提供される蛋白質素材は実用上充分な有機溶媒に対する溶解度を有しているが、更に溶解度を高めることができれば或いは該素材に有機溶媒溶解性以外の機能を付与できれば、実用範囲が大きく広まることが予測される。
【０００９】
【課題を解決するための手段】本発明者は、有機溶媒可溶性蛋白質素材につき引続き研究を行なった結果、原料蛋白質として２種以上の異なる蛋白質を併用し、これらを上記の方法で架橋高分子化することにより、得られる有機溶媒可溶性の蛋白質素材の機能（例えば有機溶媒に対する溶解性や後記する吸着性）を向上させたり、該素材の収量を増加させ得ることを見い出し、ここに本発明を完成するに至った。
【００１０】即ち、本発明は、少なくとも２種の蛋白質、水及び架橋剤を混合、架橋させ、次いで得られる蛋白質の架橋物にアルキル化剤、シッフ化剤及び酸から選ばれた少なくとも１種を反応させることを特徴とする機能性蛋白質素材の製造法に係る。
【００１１】本発明の機能性蛋白質素材（以下「本素材」という）の製造に当っては、まず２種以上の蛋白質と水と架橋剤とを混合（好ましくは激しく混合）する。これにより蛋白質と架橋剤を付加重合させて蛋白質の架橋物を得る。この時、予め蛋白質と水とを混合して蛋白質水溶液を製造し、これに架橋剤を加えてもよい。また架橋剤を有機溶媒に溶解させ、この溶液と蛋白質と水とを混合してもよい。その際、混合後の液は水相と油相とに分離し、蛋白質の架橋物の大部分は水相中に含まれる。
【００１２】本発明で使用する蛋白質としては特に制限されないが、その中でも遊離アミノ基を有する蛋白質及びペプチド類が好ましく、例えば、鶏、うずら、あひる、ガチョウ等の卵の卵白蛋白質、ホエー蛋白質、血清蛋白質（血清アルブミン等）、カゼイン、ゼラチン等を挙げることができ、これらから少なくとも２種を選択すればよい。蛋白質の使用量（２種以上の蛋白質の使用合計量）は特に制限されず広い範囲から適宜選択できるが、蛋白質濃度が通常１～５重量％程度となるように蛋白質を使用するのが好ましい。また２種以上の蛋白質を併用する場合のこれら蛋白質の併用割合は、特に制限さ

れず、得ようとする素材の機能、使用目的等に応じて適宜選択すればよいが、例えば卵白とゼラチンとを併用し、後記する物質吸着性に優れた素材を得ようとする場合には、ゼラチンを通常全量（全蛋白質使用量）の１～３０重量%程度、好ましくは５～１５重量%程度使用すればよい。

【００１３】蛋白質に反応させる架橋剤は、蛋白質間の遊離のアミノ基及びアルコール性水酸基と反応して、尿素結合、ウレタン結合、酸アミド結合等の化学的結合により蛋白質間を架橋することができ、且つ蛋白質の疎水度をコントロールできるものであれば特に制限されないが、例えば、ジイソシアネート化合物、ジアルデヒド化合物、ジケトン化合物等を挙げることができる。その中でもジイソシアネート化合物が反応性に富んでいるため、好ましく使用できる。ジイソシアネート化合物としては従来公知のものを広く使用でき、例えば、トルエンジイソシアネート（ＴＤＩ）、ジイソシアン酸ジフェニルメタン（ＭＤＩ）、ヘキサメチレンジイソシアネート（ＨＤＩ）、イソホロンジイソシアネート（ＩＰＤＩ）、ナフタリンジイソシアネート（ＮＤＩ）等の１分子中に２個以上のイソシアナート基を有する化合物を挙げることができる。架橋剤の使用量は特に制限されないが、通常は反応させる蛋白質の一次構造からアミノ基及びアルコール性水酸基の総モル数を算出し、それに応じて架橋剤の使用量（モル数）を決定すればよい。

【００１４】架橋剤を有機溶媒に溶解させる場合に使用する有機溶媒としては、蛋白質と架橋剤の界面付加重合を可能にする公知の有機溶媒を広く使用でき、例えばクロロホルム、ヘキサン、トルエン等を挙げることができる。

【００１５】蛋白質と架橋剤との反応は、通常蛋白質と水と架橋剤とを混合することにより行なわれる。この反応の条件は、蛋白質間に架橋が起こる条件であれば特に制限されないが、通常室温下に１時間以上程度行なえばよい。

【００１６】上記反応により生成する蛋白質の架橋物は、通常の分離精製手段に従って反応混合物中から単離して次の反応に供してもよく、或いは蛋白質の架橋物を含む反応混合物をそのまま次の反応に供してもよい。

【００１７】次いで蛋白質の架橋物とアルキル化剤、シッフ化剤及び酸から選ばれた少なくとも１種を反応させることにより、本素材が生成する。

【００１８】まずアルキル化剤を加える場合につき説明する。アルキル化剤の添加により、蛋白質の架橋物中の残存アミノ基、フェノール性水酸基及びカルボキシル基がアルキル化（修飾）され、目的とする本素材をゲル状物として得られるものと推定される。アルキル化剤としては公知のものを広く使用でき、例えば、ジメチル硫酸等のジアルキル硫酸、硫酸アルキル、ハロゲン化アルキル、スルホン酸アルキル等を挙げることができる。尚、

ジアルキル硫酸、硫酸アルキル等を使用すると、アルキル化剤としての効果と共にｐＨ調整剤としての効果もある。アルキル化剤の添加量は上記架橋反応の程度等に応じて適宜選択すればよいが、通常はアミノ基が全て架橋していると仮定し、反応させる蛋白質の一次構造からフェノール性水酸基及びカルボキシル基の総モル数を算出し、それに応じて架橋剤の使用量（モル数）を決定すればよい。得られるゲル状物は、そのまま本素材として使用可能であるが、遠心分離、濾過等の通常の分離手段で水分を除去し、必要に応じて水洗した後、真空式ベルト乾燥機やフリーズドライ機等で乾燥し、粉末品として使用してもよい。

【００１９】次に、シッフ化剤を加える場合につき説明する。シッフ化剤は蛋白質架橋物中の残存アミノ基とシッフ化反応してアミノ基を修飾する。シッフ化剤の添加により、アルキル化剤の場合と同様に本素材をゲル状物として得ることができる。シッフ化剤としては、従来から公知のもの、例えばアルデヒド類等を挙げることができる。得られるゲル状物は、上記と同様に分離、乾燥できる。

【００２０】更に酸を加える場合につき説明する。水相に酸を加え、該相のｐＨを原料蛋白質の等電点以下に調整する。これにより、有機溶媒に可溶で且つ水に不溶の本素材が沈殿する。得られる沈澱物は上記と同様の分離手段及び乾燥手段により、粉末化することができる。酸としては公知のものをいずれも使用でき、例えば、塩酸、クエン酸、琥珀酸、酢酸、乳酸、酒石酸、フマル酸、リンゴ酸、アジピン酸、グルコノデルタラクトン、グルコン酸、アスコルビン酸、レブリン酸、フタル酸等を挙げることができる。尚、架橋が不充分であったり疎水性が不足すると、酸を加えてｐＨを下げても沈澱物は析出するが、本素材が生成しない虞れがある。従って、酸を単独で使用する場合は、前工程の架橋剤の使用量を通常の２倍程度以上、好ましくは２～３倍程度とするのがよい。

【００２１】上記アルキル化剤、シッフ化剤及び酸はそれぞれ単独で使用できるが、これらを２者又は３者併用しても差し支えない。

【００２２】本発明においては、架橋反応等の反応を有利に進行させたり、反応速度を早めたり、本素材の性能や収率を向上させたりすることを目的として、例えば、蛋白質と架橋剤とを反応させるのに先立ち蛋白質に前処理を施してもよい。

【００２３】上記蛋白質の前処理方法としては特に制限されないが、例えば、希釈、電気透析、加熱、ｐＨ調整、遠心分離、濾過等であり、これらの中の１種を行なってもよく、或いは２種以上を適宜組合わせて行なってもよい。より具体的には特願平２－４１８８７６号、特願平４－５３４６６号等に記載されている。

【００２４】希釈は主として水を用いて行ない、例えば

卵白蛋白質で説明すると、その希釈倍率は通常２倍以上、好ましくは２～５倍程度、より好ましくは２～３倍程度とするのがよい。希釈に用いられる水としては、例えば脱イオン水、蒸留水、純水、水道水等が挙げられる。

【００２５】電気透析は、蛋白質又はその希釈物のイオン濃度を下げるために行なわれる。電気透析は常法に従って行ない得る。尚、電気透析を行なうのに先立ち、蛋白質又はその希釈物のｐＨを調整しておくのが好ましい。この場合には、蛋白質又はその希釈物のｐＨを酸性から中性域（通常ｐＨ５～８程度、好ましくは６～７程度）にするのがよい。斯かるｐＨの調整には、酸が用いられる。添加されるべき酸としては、特に限定されないが、食品添加物中の酸味料となるものが好適である。斯かる酸としては、具体的にはクエン酸、琥珀酸、酢酸、乳酸、酒石酸、フマル酸、リンゴ酸、アジピン酸、グルコノデルタラクトン、グルコン酸、アスコルビン酸、塩酸等を例示できる。

【００２６】加熱は特に限定されないが、通常約７５℃以上で約３０分前後又はそれ以上で行なわれる。尚、加熱する前の卵白又はその希釈物のｐＨはアルカリ域（通常ｐＨ８以上、好ましくは９以上）にあるのが好ましい。従って、ｐＨが前記範囲よりも低い時、特に電気透析を行なってｐＨが酸性乃至中性域にある場合には、適当なアルカリ剤を用いて前記ｐＨ域に調整してもよい。アルカリ剤としては、従来公知のものを広く使用でき、例えば水酸化ナトリウム、炭酸カルシウム、炭酸アンモニウム、炭酸ナトリウム、炭酸水素アンモニウム、炭酸カリウム、炭酸水素ナトリウム、炭酸マグネシウム、ポリリン酸等を挙げることができる。

【００２７】上記の希釈、電気透析、加熱等の各処理により凝集物や沈殿物が析出する場合には、これらを通常の分離手段で除去するのが好ましい。例えばフィルター濾過したり、遠心分離すればよい。

【００２８】また蛋白質と架橋剤とを反応させる際には、蛋白質又はその希釈物のｐＨはアルカリ域（通常ｐＨ８以上、好ましくは１０～１２程度）にあるのが好ましい。従って、ｐＨが前記範囲よりも低い場合には、適当なアルカリ剤を加えて前記ｐＨ域に調整してもよい。

【００２９】更に蛋白質の架橋物とアルキル化剤やシッフ化剤とを反応させるに先立ち、ｐＨ調整を行なってもよい。

【００３０】以下に本素材を得るための好ましい実施態様の一例を挙げる。

【００３１】（ａ）　卵白又はその水希釈物（卵白濃度１～５重量％程度）のｐＨを酸性乃至中性域に調整し、電気透析によりイオン強度を下げ、得られる液のｐＨを９．０以上に調整し、加熱する。

【００３２】（ｂ）　加熱された卵白水溶液に他の蛋白質を添加、混合し、蛋白質混合水溶液を調製する。

【００３３】（ｃ）　加熱された蛋白質混合水溶液のｐＨをアルカリ性域（例えばｐＨ１０～１２）に調整する。

【００３４】（ｄ）　ｐＨ調整後の蛋白質混合水溶液と架橋剤の有機溶媒溶液を混合して激しく撹拌して静置し、水相と油相に分離させる。

【００３５】（ｅ）　水相（通常ｐＨ６．５～９程度）を分取し、更にアルカリ性域（例えばｐＨ１２程度）にｐＨ調整する。

【００３６】（ｆ）　更に撹拌下アルキル化剤を加え、析出するゲル状物を遠心分離により分離し、水で洗浄し、凍結乾燥する。

【００３７】斯くして得られる本素材は、アミノ酸分析の結果から、蛋白質であることが確認される。本素材の最大の特徴は、本素材が蛋白質であるにもかかわらずジメチルホルムアミド（ＤＭＦ）、ジメチルスルホキシド（ＤＭＳＯ）、ジメチルアセトアミド（ＤＭＡ）、フェノール、蟻酸等の有機溶媒に溶解し水に溶解しない点である。ところが本素材を水に分散させてｐＨをアルカリ側（およそ６以上）に調整すると、有機溶媒に不溶で水溶性の素材に変化する。而して再度その水溶液のｐＨを本素材の等電点以下に調整すると、有機溶媒可溶且つ水不溶の素材に変化する。この変化は、前記の如きｐＨ調整により何度でも可逆的に起こる。また本素材は極めて優れた物質吸着能を有している。

【００３８】

【発明の効果】

（１）　本発明によれば、２種以上の原料蛋白質を併用することにより、得られる有機溶媒溶解性の本素材の機能を向上させたり、或いは本素材の収率を高めたりすることができる。例えばゼラチンと卵白とを併用すると、得られる本素材にゼラチンの保護コロイド性が付与されると共に、卵白単独使用の場合に比し、収率が大幅に向上する。

【００３９】（２）　本発明によれば、特願平４－５３４６６号に記載のものよりも更に有機溶媒に対する溶解性に優れた蛋白質素材を提供できる。該素材と合成樹脂とを有機溶媒に溶解混合して溶媒を除くと、該素材は樹脂中に極めて均一に分散するので、樹脂に蛋白質の微粉末を混合する場合に比べてはるかに優れた蛋白質的機能を発揮し得る。

【００４０】（３）　本素材は、優れた吸着能を有していると共に、ｐＨの変化により可逆的に物性が変化するという特徴をも有している。この性質を利用して、本素材を物質の吸脱着剤として用いることができる。この吸脱着性の利用としては、例えば、廃水からの着色や汚染の原因となる物質の分離・有用物質の回収、二酸化チタンや酸化ジルコニウムを吸着させ紫外線吸収剤として繊維や化粧品等に適用すること等が考えられる。

【００４１】（４）　本素材は有機溶媒に可溶であるた

め、プラスチックのフィルム、シート、チューブ等に非常に均一に分散させることができ、新たな機能をプラスチックに付与することができる。

【００４２】例えば、本素材を衣類用合成繊維に応用すると、プラスチックのみの繊維に比べて、天然素材のようなべと付かない肌触りが得られ、湿気の吸入性と放出性のバランス、保温性等が良好で、結露し難いという優れた性能が発現される。

【００４３】また、化粧用等のパフは従来ラテックス等の多孔性ゴムを凝固加硫するか又はウレタンフォームを成形して製造されているが、その表面のしっとり感、肌ざわり等の点で満足の行くものが得られていない。そこでウレタンフォームを製造する際に、本素材の有機溶媒溶液を添加することにより、絹に似たしっとり感、滑らかな肌ざわりを有するスポンジ（パフ）を得ることができる。尚、ウレタンフォームの製造は従来の方法に従えばよく、また本素材の添加量等は、得られるスポンジの使用目的等に応じて適宜選択すればよい。

【００４４】（５）　本素材は、その分子中に例えばカルボキシル基等の官能基に起因する性能を有していることから、種々の用途に使用できる。例えば、本素材は界面活性効果を有し、界面活性剤、繊維助剤、帯電防止剤、染料固着剤、染料助剤等として利用できる。また本素材（のカルボキシル基）とアスコルビン酸、パントテン酸等のエステルは、化粧品原料として利用できる。本素材は金属イオンを捕捉するキレート剤の原料としても使用できる。

【００４５】その他、本素材は、例えばインクジェット、記録紙、人工皮膚、人工臓器、人工皮革、分析・理化学検査用の固定化剤、農薬・肥料等のコーティング剤、Ｌ－Ｂ膜形成のバイオセンサー、人工の機能性膜（例えば人工の細胞膜等）、マイクロカプセル素材、ドラッグデリバリーシステム等の用途に応用できる。

【００４６】

【実施例】以下に実施例を挙げ、本発明を一層明瞭なものとする。

【００４７】実施例１

冷凍鶏卵白８００ｇを３５℃の温水中に放置して解凍し、３２メッシュのフィルターを通した後、水道水で２．５倍に希釈し、穏やかな撹拌下に２Ｍ－クエン酸を加え、ｐＨ６．８に調整した。この調整卵白を４２メッシュのフィルターを通し、更に遠心分離（７０００ｒｐｍ×１０分間）し、ｐＨ調整により析出物を除去した。得られた上澄液を電気透析（電気透析：ＣＳ－Ｏ型・商品名・旭硝子（株）製、流量：２５０リットル／ｈｒ．、定電圧：１４Ｖ）、電導度を９５０μＳ／ｃｍとした。この透析液を１Ｎ及び６Ｎの水酸化ナトリウムを加え、ｐＨ１０に調整した。これを沸騰水中で３０分間加熱し、室温まで冷却し、１００メッシュのフィルターを通した卵白液を得た。この卵白液の吸光度は０．３７０（２８０ｎｍ）であり、卵白濃度は約４％であった。

【００４８】得られた卵白液８００ｍｌにゼラチン〔新田ゼラチン（株）製、分子量１５０００〕３ｇを溶解させ、イオン水を加えて１リットルとし、１Ｎ及び６Ｎの水酸化ナトリウムを加え、ｐＨ１２に調整した。この混合液を４５℃に加温した。一方１８．４ｇのトルエンジイソシアネートをクロロホルム３００ｇに混ぜた後、これを加温中の混合液に入れて２時間撹拌を続けた後、室温で放置し、水相とクロロホルム相に分離した。水相部を遠心分離（８０００ｒｐｍ×１０分）により分取した。

【００４９】この上澄液をクエン酸でｐＨ４に調整すると本素材が沈殿するので、再度遠心分離（１００００ｒｐｍ×１５分）した。分取した本素材はＤＭＦに易溶であり、この液を水に入れると蛋白質の析出が認められた。この沈殿物を凍結乾燥して得られた粉末もＤＭＦに可溶であり、この液も水に入れると蛋白質の析出が認められた。

【００５０】実施例２

実施例１と同様にして、卵白量の５～２０重量％のゼラチンを加え、本素材を製造した。

【００５１】得られた本素材４ｇにイオン水を加えて全量１リットルとし、撹拌下に１Ｎ及び６Ｎの水酸化ナトリウムを加えてｐＨ７に調整して本素材を水に溶解させた。この溶解液にパプリカ色素〔天然パプリカ色素の乳化調合液、商品名：パプリカベース１５０、三栄化学工業（株）製〕１ｍｌを添加、混合し、２Ｍクエン酸を添加してｐＨ４に調整し室温で静置すると、パプリカ色素を吸着した本素材が析出した。２時間室温で放置後、上澄液の吸光度（４７０ｎｍ）を測定し、パプリカ色素の残量を調べた。結果を表１に示す。

【００５２】比較のため、ゼラチンを加えない以外は実施例１と同様にして機能性蛋白質素材（即ち特願平４－５３４６６号に記載の素材）を得、上記と同様に処理してハムプリカ色素の残量を調べた。結果を表１に併せて示す。

【００５３】

【表１】

対鶏卵白のゼラチン量（%）	吸光度
0	0．52
5	0．31
10	0．26
20	0．14

【００５４】表１からゼラチンの添加量が多い程、パプリカ色素の残量が少ない（吸光度が低い）ことが判る。従って、ゼラチンの保護コロイド性が発揮されて、ゼラチンの増加に伴い吸着能が高くなることが推測される。即ち、本方法によれば、ゼラチン本来の機能を保持したまま有機溶媒に可溶という機能を有する蛋白質素材が得

られる。

【００５５】実施例３

実施例１で得られた本素材２．５ｇにイオン水を加えて全量１リットルとし、１Ｎ及び６Ｎの水酸化ナトリウムを加えてｐＨ７に調整して本素材を水に溶解させた。この溶解液に二酸化チタン〔商品名：ＴＡＦ－１００、粒子径０．０５μｍ、富士チタン工業（株）製〕１．５ｇを添加し、超音波処理及びディスパーサー処理を各１０分行なって本素材及び二酸化チタンを分散させた後、ｐＨ４に調整して二酸化チタンを吸着した本素材を析出、沈殿させた。これを遠心分離（１００００ｒｐｍ×２０分）し、二酸化チタンを吸着した本素材を分取した。

【００５６】尚、超音波発生機としては商品名「ＢＲＡＮＳＯＮ　２２００」〔ヤマト科学（株）製〕を、またディスパーサーとしては商品名「ウルトラディスパーサーＭｏｄｅｌ　ＬＫ－２２」〔ヤマト科学（株）製〕をそれぞれ用いた。

【００５７】二酸化チタンを吸着した本素材をイオン水に分散させ、２００～４００ｎｍの紫外部吸収を測定した。結果を図１に示す。図１から、二酸化チタン吸着の本素材が二酸化チタン自体と同様の紫外部吸着スペクトルを示すことが判る。従って、本素材に二酸化チタンが吸着していることが明らかである。

【００５８】実施例４

ナイロン６６チップを蟻酸に１日要して溶解し、２０％ナイロン－蟻酸溶液（Ａ液）を調製した。また、実施例１で得られた本素材を蟻酸に溶解し、３％機能性蛋白質－蟻酸溶液（Ｂ液）を調製した。

【００５９】Ａ液１０ｇにＢ液２ｍｌを添加して均一になるように撹拌し、約３％の本素材を含むナイロン溶液を調製した。このナイロン液を平らなガラス板上に延ばし風乾して溶媒を飛散させると、白色の引張り力の弱い膜が生成した。

【００６０】この白色膜を、インパルスシーラー〔商品名：インパルスシーラーＦ１－２００－１０ｗ型〕で加熱加圧したところ、透明性で引張り強度のあるナイロンフィルムが得られた。このものは、ナイロン単品にはない透湿性が付与されていた。

【００６１】実施例５

冷凍鶏卵白８００ｇを３５℃の温水中に８時間放置して＊解凍し、３２メッシュのフィルターで濾過した後、水道水で２．５倍に希釈し、穏やかな撹拌下に２Ｍ－クエン酸を加え、ｐＨ６．８に調整した。この調整卵白を４２メッシュのフィルターを通し、更に遠心分離（７０００ｒｐｍ×１０分間）し、ｐＨ調整による析出物を除去した。

【００６２】得られた上澄液を電気透析（電気透析：ＣＳ－Ｏ型・商品名・旭硝子（株）製、流量：２５０リットル／ｈｒ．、定電圧：１４Ｖ）し、電導度を９８５μＳ／ｃｍとした。この透析液に１Ｎ及び６Ｎの水酸化ナトリウムを加え、ｐＨ１０に調整した。これを沸騰水中で３０分間加熱し、室温まで冷却し、２００メッシュのフィルターで濾過し、吸光度が０．３８６（２８０ｎｍ）の卵白液を得、これを希釈してｐＨ１２に調整した。この希釈卵白液の吸光度は０．１６３（２８０ｎｍ）であった。

【００６３】上記で得られた希釈卵白液４００ｍｌにホエー蛋白質〔商品名：ＤＦ－ＷＰＣ、明治乳業（株）製、部分脱脂ホエー蛋白質〕７ｇを溶解させ、再度ｐＨを１２に調整した。この時の吸光度は０．３２８（２８０ｎｍ）であった。この混合液を４５℃に加温し、これにトルエンジイソシアネート７ｇとクロロホルム３００ｇとの混合物を加えて２時間撹拌した後、室温で放置して水相とクロロホルム相とに分離させ、水相を分取した。この水相部にクエン酸を加えてｐＨ４に調整すると本素材が析出、沈殿するので、遠心分離（１００００ｒｐｍ×３０分）して本素材を分取した。

【００６４】本素材はＤＭＦに易溶であり、この液を水に入れると蛋白質の析出が認められた。本素材を凍結乾燥して得られた粉末もＤＭＦに可溶であり、この液を水に入れると蛋白質の析出が認められた。

【００６５】実施例６

実施例１で得られた鶏卵白とゼラチンとを原料とする本素材、実施例５で得られた鶏卵白とホエー蛋白質を原料とする本素材及び鶏卵白のみを原料とする素材（実施例２）の溶媒（ＤＭＦ）に対する溶解性を調べた。結果を表２に示す。

【００６６】

【表２】

素材の原料	素材濃度	溶解性
鶏卵白＋ホエー蛋白質	４０％	溶解後２日で白濁ゲル化
鶏卵白＋ゼラチン	２０％	溶解後２日で白濁ゲル化
鶏卵白	２０％	溶解後２時間で白濁ゲル化

【００６７】表２から、原料として２種の蛋白質を併用することにより、得られる蛋白質素材の有機溶媒に対する溶解度が大きくなることが判る。

【００６８】実施例７

実施例５で得られた本素材４ｇにイオン水を加えて全量１リットルとし、撹拌下に１Ｎ及び６Ｎの水酸化ナトリウムを加えてｐＨ７に調整して本素材を溶解させた。この溶解液にパプリカ色素１ｍｌ〔パプリカベース１５０〕を添加、混合し、２Ｍクエン酸を添加してｐＨ３．３に調整し室温で静置すると、パプリカ色素を吸着した

本素材が析出した。

【００６９】実施例８

冷凍鶏卵白８００ｇを３５℃の温水中に８時間放置して解凍し、３２メッシュのフィルターで濾過し、水道水で４倍に希釈し、遠心分離（７０００ｒｐｍ×１０分間）し、ｐＨ１２に調整した。この希釈卵白液４００ｍｌにホエー蛋白質〔商品名：ＤＦ－ＷＰＣ、部分脱脂ホエー蛋白質、明治乳業（株）製〕７ｇを溶解させ、再度ｐＨを１２に調整した。この時の吸光度は０．３５５（２８０ｎｍ）であった。

【００７０】この蛋白混合液を４５℃に加温し、これにトルエンジイソシアネート７ｇとクロロホルム３００ｇとの混合物を加えて２時間攪拌した後、室温で放置して水相とクロロホルム相とに分離した。分取した水相を遠心分離（８０００ｒｐｍ×１０分）して不純物を除去した後、２Ｍ－クエン酸を加えてｐＨを３．５まで下げると本素材が析出沈殿するので、遠心分離（８０００ｒｐｍ×１０分）して本素材を分取した。本素材はＤＭＦに易溶であり、水不溶性であった。

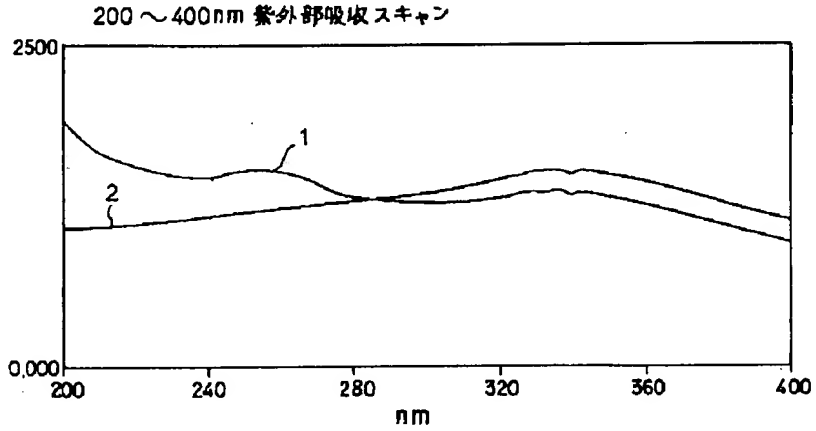
【００７１】実施例９

実施例８におけるトルエンジイソシアネート７ｇとクロロホルム３００ｇの混合物の代りにトルエンジイソシアネート１０ｇを使用する以外は、実施例８と同様にしてＤＭＦに易溶で且つ水不溶性の本素材を得た。

【図面の簡単な説明】

【図１】本素材と二酸化チタンの凝集物の２００～４００ｎｍの紫外部吸収スペクトルを示す図面である。

【図１】



1　凝集物：TiO_2＝5：1

2　TiO_2 のみ

フロントページの続き

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PROCESS FOR THE PRODUCTION OF FUNCTIONAL PROTEIN MATERIAL

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PROCESS FOR THE PRODUCTION OF FUNCTIONAL PROTEIN MATERIAL

[Kinousei tanpakushitsu sozaino seizou houhou]

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[There are no amendments to this patent.]

Claims

/2*

1. A process for the production of a functional protein material characterized by mixing at least 2 kinds of proteins, water and cross-linking agent to carry out cross-linking and

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* [Numbers in margin indicate pagination of the original text.]

subsequently, allowing the prepared cross-linked product of the proteins to react with at least one kind of compound selected from alkylation agents, Schiff's reagents and acids.

2. The process for the production of a functional protein material of Claim 1, wherein the protein is a protein or peptide having a free amino group.

3. The process for the production of a functional protein material of Claim 2, wherein the protein or peptide having a free amino acid is at least one kind of substance selected from egg white protein of chicken, quail, duck or goose, whey protein, serum albumin and casein.

Detailed explanation of the invention

[0001]

Industrial application field

This invention pertains to a process for the production of a functional protein material. In particular, it pertains to a process for the production of a functional protein material soluble in an organic solvent.

[0002]

Prior art and problems

In order to improve skin touch feel, moisture release and absorption balance, heat retention, etc., of synthetic fibers there have been attempts to add proteins to synthetic fibers.

[0003]

For example, Japanese Patent Kokai Application No. Hei 1[1989]-293143 discloses a fiber of a synthetic resin with dispersed gelatin and silk micropowder, but because of particle size variations of the micropowders used, it is not uniformly dispersed in the resin, and a synthetic fiber prepared from this synthetic resin is not necessarily satisfactory with respect to skin touch, moisture absorption and release balance, heat retention, etc. In addition, there are very difficult handling problems with gelatin and silk micropowders such as complex pulverization procedures, scattering and flying of pulverized particles, etc.

[0004]

Preferable results to improve the dispersibility of proteins in resins are apparently obtainable by using a protein which can be dissolved in an organic solvent which also can dissolve the resin used.

[0005]

As a protein soluble in an organic solvent, for example, a simple protein called prolamin has been known, but it is only soluble in 60-90% ethanol (that is, aqueous organic solvent), but it is insoluble in ethanol of 90% or higher purity or in other organic solvents, and consequently, its practical application is difficult.

[0006]

Furthermore, Japanese Kokai Patent Application No. Sho 52[1977]-25800 discloses that a modified protein prepared by carrying out hydrolysis of an adduct of a protein and isocyanate compound in the presence of an organic solvent or aqueous organic solvent is soluble in dimethylsulfoxide (DMSO), dimethylacetamide (DMA), etc. However, such a protein is a so-called denatured protein with a low molecular weight and a side chain having amide bonding and urethane bonding severed, showing infrared absorption 1740 cm^{-1} and 1222 cm^{-1}, and only side chain having urea bonding remaining, and consequently, there is a risk of the protein function being reduced. In addition, the solubility of such a modified protein in an organic solvent is not necessarily sufficient.

[0007]

Problems to be solved by the invention

The inventors of this invention studied diligently by considering the current situation of the prior art as described above, and as a result they found that a protein material soluble in an organic solvent could be prepared by forming a cross-linked polymer of the raw material protein by using a specific cross-linking agent such as diisocyanate, etc., and they previously applied for a patent (Japanese Patent Application No. Hei 4[1992]-53466).

[0008]

The protein material of the invention of the above patent application has a practically satisfactory solubility in an organic solvent, but if it is possible to improve its solubility further or add functions other than dissolution in an organic solvent to the material, the practical application range of this material is expected to be expanded.

0009

Means to solve the problems

The inventors assiduously researched to discover an organic solvent-soluble protein material, as a result, they found that an organic solvent-soluble protein material prepared by using 2 or more kinds of proteins as the raw material protein and carrying out the method

described above to form a cross-linked polymer showed improved functions (such as solubility in an organic solvent and adsorption explained later) as well as improved yield of the material, and they arrived at the present invention.

[0010]

Specifically, this invention pertains to a process for the production of a functional protein material characterized by mixing at least 2 kinds of proteins, water and cross-linking agent to carry out cross-linking and subsequently, allowing the cross-linked product of the proteins prepared to react with at least one kind of compound selected from alkylation agent, Schiff's reagent and acid.

[0011]

To produce the functional protein material of this invention (called "material of this invention" below), two or more kinds of proteins are mixed (preferably vigorously) with water and a cross-linking agent first. As a result, the addition polymerization of the cross-linking agent to the proteins is carried out giving cross-linked proteins, In this case, the proteins and water may be mixed in advance to obtain an aqueous solution of proteins, to which the cross-linking agent may be added. Furthermore, the cross-linking agent may be dissolved in an organic solvent, and this solution, proteins and water may be mixed. In this case, the reaction mixture after mixing separates to aqueous and oil phases, and the cross-linked proteins are mostly found in the aqueous phase.

[0012]

The proteins usable in this invention are not especially restricted, but among those proteins, proteins and peptides having free amino groups are preferable. For example, there are egg white protein of chicken, quail, duck, goose, etc., whey protein, blood serum protein (serum albumin, etc.), casein, gelatin, etc., and at least 2 kinds of proteins may be selected from them. The amount of proteins to be used (total amount of 2 or more kinds of proteins) is not especially restricted, and it is suitably selectable from a wide range, but the proteins are preferably used so that the protein concentration is generally in the range of about 1-5 wt%. The proportion of 2 or more kinds of proteins is not especially restricted, and it is suitably selected depending on the function of the material to be prepared, application purpose, etc., but, for example, in the case of egg white and gelatin concomitantly used to obtain a material having excellent substance-adsorption property, the amount of gelatin is in the range of 1-30 wt%, preferably 5-15 wt% to the total amount (total amount of proteins used). /3

[0013]

The cross-linking agent allowed to react with proteins is not especially restricted as long as it enables proteins to be cross-linked through chemical bonds such as urea bonding, urethane bonding, acid amide bonding, etc., by carrying the reaction with free amino and alcoholic hydroxyl groups between proteins and can control the degree of hydrophobicity of proteins, but the use of, for example, diisocyanate, dialdehyde, diketone compounds, etc., is preferable. Among these compounds, the use of diisocyanate compounds is optimal because of rich reactivity. As a diisocyanate compound, those previously known compounds are usable, and there are, for example, those compounds having 2 or more isocyanate groups per molecule such as toluene diisocyanate (TDI), diphenylmethane diisocyanulate (MDI), hexamethylene diisocyanate (HDI), isophorone diisocyanate (IPD), naphthalene diisocyanate (NDI), etc. The amount of this cross-linking agent to be used is not especially restricted, but in general, the amount (number of moles) of the cross-linking agent to be used is determined depending on the number of total moles of amino and alcoholic hydroxyl groups calculated from the primary structures of proteins.

[0014]

As an organic solvent if the cross-linking agent is used after dissolving in an organic solvent first, there are known organic solvents enabling interfacial addition polymerization of the cross-linking agent and proteins, and specific examples include chloroform, hexane, toluene, etc.

[0015]

The reaction between the cross-linking agent and proteins is generally carried out by mixing the cross-linking agent, proteins and water. The reaction conditions to be employed in this case are not especially limited as long as the cross-linking reactions of the proteins can be carried out, but in general, the reaction is carried out at a room temperature for 1 hour or longer.

[0016]

The cross-linked products of the proteins prepared by the above reaction are isolated from the reaction mixture by using conventional isolation and purification procedures and used for the subsequent reaction. Alternatively, the reaction mixture containing the cross-linked products may be used as is for the subsequent reaction.

[0017]

The cross-linked product of the proteins is allowed to react with at least one kind of compound selected from alkylation agents, Schiff's reagents and acids to obtain the material of this invention.

[0018]

The case of adding an alkylation agent is explained below. By adding an alkylation agent, the residual amino, phenolic hydroxyl and carboxyl groups in the cross-linked product of proteins are alkylated (modified) to obtain the desired material of this invention in a gel state. As an alkylation agent, any of those previously known alkylation agents may be used, and specific examples include dialkylsulfuric acids such as diemthylsulfuric acid, etc., alkyl sulfate, alkyl halide, alkyl sulfonate, etc. Incidentally, if dialkylsulfuric acid, alkyl sulfate, etc., are used, there are effects as a pH adjuster in addition to those as an alkylation agent. The amount of the alkylation agent to be added is suitably selected depending on the extent of the above cross-linking agent, etc., but in general, the amount (number of moles) of the cross-linking agent [sic] to be used is determined depending on the number of total moles of carboxyl and phenolic hydroxyl groups calculated from the primary structures of proteins. The gel prepared is usable as is, or alternatively, water is removed by using a conventional method such as centrifugation, filtration, etc., and after washing with water, if necessary, it is dried by using a vacuum belt dryer, freeze-drying machine, etc., to obtain a powder product.

[0019]

The reaction with a Schiff's reagent is explained as follows. The Schiff's reagent can modify amino groups by carrying out Schiff's reaction with the residual amino groups of the cross-linked product of proteins. As a result of the addition of this Schiff's reagent, the material of this invention is obtained in its gel state as in the case of the addition of an alkylation agent. As a Schiff's reagent, previously known compounds such as aldehydes, etc., are usable. The gel prepared is isolated and dried as described above.

[0020]

Furthermore, the addition of an acid is explained as follows. The acid is added to the aqueous phase to adjust the pH of the phase below the isoelectric point of the raw material protein. As a result, the material of this invention soluble in an organic solvent but insoluble in water is precipitated. The precipitates obtained are made into a powder by using the same isolation and drying procedures as those described above. As an acid, any of the known acids are usable, and specific examples include hydrochloric acid, citric acid, succinic acid, acetic acid,

lactic acid, tartaric acid, fumaric acid, malic acid, adipic acid, gluconodeltalactone, gluconic acid, ascorbic acid, levulinic acid, phthalic acid, etc. Incidentally, if the cross-linking is insufficient or the hydrophobic property is poor, precipitates are formed by lowering the pH with the acid, but there is a risk of the material of this invention not being formed. Therefore, if an acid alone is used, the amount of the cross-linking agent used in the prior stage is increased to 2 times or more, preferably 2-3 times the conventional amount.

[0021]

Those alkylation agents, Schiff's reagents and acids may be used alone or as a mixture of 2 or 3 kinds.

[0022]

In this invention, the proteins may, for example, be pretreated before carrying out the reactions between these proteins and a cross-linking agent for the purpose of carrying out the reactions such as cross-linking reaction, etc., advantageously, increasing the rate of reaction or improving the yield of the reaction or the performance of the material of this invention.

[0023]

The method for the above pretreatment usable in this invention for those proteins is not especially restricted, for example, there are treatments such as dilution, electrodialysis, heating, pH adjustment, centrifugation, filtration, etc., which may be carried out alone or in combination of 2 or more kinds. Specific examples are found in the Japanese Patent Application Nos. Hei 2[1990]-418876, Hei 4[1992]-53466, etc.

[0024]

The dilution is mostly carried out by using water. For example, in the case of egg white protein, the rate of dilution is generally 2 times or more, preferably 2-5 times and optimally 2-3 times. As specific examples of water to be used for this dilution, there are deionized water, distilled water, purified water, tap water, etc. /4

[0025]

The electrodialysis is carried out to lower the ion concentration of the proteins or diluted solution, and it is carried out using conventional procedures. Incidentally, prior to this electrodialysis, the pH of the proteins or their diluted solution is preferably adjusted. In this case, the pH of the proteins or their diluted solution is in the range of acidic to neutral (generally pH 5-8, preferably pH 6-7). This pH adjustment is carried out by using an acid. The acid usable in this

case is not especially limited, but is suitably one capable of becoming an agent providing food additives with an acidic taste. Specifically, there are, for example, citric acid, succinic acid, acetic acid, lactic acid, tartaric acid, fumaric acid, malic acid, adipic acid, gluconodeltalactone, gluconic acid, ascorbic acid, hydrochloric acid, etc.

[0026]

The heating conditions are not especially restricted, but it is generally carried out at a temperature above about 75°C for about 30 min or longer. Incidentally, the pH of egg white or its diluted solution is preferably in a basic range (generally pH 8 or higher, preferably 9 or higher). Therefore, if the pH is lower than this range, especially in the case of acidic or neutral range pH after carrying out electrodialysis, a suitable base may be used to adjust the pH to the above range. Any of those previously known bases is usable for this, and specifically, there are, for example, sodium hydroxide, calcium carbonate, ammonium carbonate, sodium carbonate, ammonium hydrogen carbonate, potassium carbonate, sodium hydrogen carbonate, magnesium carbonate, polyphosphoric acid, etc.

[0027]

If any aggregates or precipitates are formed in the above treatments such as dilution, electrodialysis, heating, etc., they are preferably removed by using a conventional separation means such as filtration or centrifugation.

[0028]

Furthermore, when the reaction of proteins and a cross-linking agent is carried out, the pH is desirably in a basic range (generally pH 8 or higher, preferably in the range of 10-12). Therefore, if the pH is lower than this range, the pH is preferably adjusted by using a suitable base.

[0029]

Furthermore, prior to carrying out the reaction of the cross-linked protein product with an alkylation agent, Schiff's reagent or acid, the pH adjustment may be carried out.

[0030]

An example of the preferred embodiment of this invention to obtain the material of this invention is mentioned as follows.

[0031]

(a) The pH of egg white or its diluted aqueous solution (egg white concentration of about 1-5 wt%) is adjusted in an acidic or neutral range, the ionic strength is reduced by carrying out electrodialysis, the pH of the solution obtained is raised above 9.0 and subsequently heated.

[0032]

(b) To the heated aqueous solution of egg white, other proteins are added and mixed to obtain an aqueous protein solution mixture.

[0033]

(c) The pH of the heated aqueous protein solution mixture is adjusted in a basic range (e.g., pH 10-12).

[0034]

(d) The aqueous protein solution mixture after pH adjustment is mixed with an organic solvent solution of a cross-linking agent, the mixture is shaken vigorously, and subsequently it is allowed to stand for separation into aqueous and oil phases.

[0035]

(e) The aqueous phase (generally pH 6.5-9) is isolated, and its pH is adjusted to a basic range (such as pH 12).

[0036]

(f) Furthermore, an alkylation agent is added wile stirring, the gel precipitated is isolated by centrifugation, washed with water and freeze-dried.

[0037]

The material of this invention prepared as described above has been confirmed to be a protein as a result of amino acid analysis. One of the most significant characteristics of this invention is found in a point that the material of this invention is soluble in organic solvents such as dimethylformamide (DMF), dimethylsulfoxide (DMSO), dimethylacetamide (DMA), phenol, formic acid, etc., in spite of being a protein and insoluble in water. However, if the material of this invention is dispersed in water, and the pH is changed to a basic range (above about 6), it is changed to become insoluble in an organic solvent and soluble in water. If the pH of the aqueous solution is adjusted below the isoelectric point of the material of this invention, it is changed to a material soluble in an organic solvent and insoluble in water. The changes are reversible many

times by the above pH adjustment. Furthermore, the material of this invention also has an excellent ability for substance adsorption.

[0038]

Effect of the invention

(1) According to this invention, the concomitant use of two or more kinds of proteins enables improvement in the function of the prepared organic solvent-soluble material of this invention in the yield of the material of this invention. For example, if gelatin and egg white are concomitantly used, the prepared material of this invention is provided with a protective colloidal property of gelatin, and at the same time, the yield is drastically improved from that with egg white used alone.

[0039]

(2) This invention provides a protein material having a solubility in an organic solvent better than that described in the Japanese Kokai Patent Application No. Hei 4[1992]-53466. If the material and a synthetic resin are dissolved in an organic solvent, and the solvent is removed, the material is dispersed uniformly throughout the resin, and thus, it provides an excellent protein function far better than mixing the resin with protein micropowder.

[0040]

(3) The material of this invention also has an excellent ability for adsorption and at the same time, characteristics of reversible physical properties depending on pH changes. This property may be utilized to use the material of this invention as an adsorbent. The adsorption-desorption properties allow applications such as isolation of substances causing coloration, contamination, etc., and recovery of useful substances from waste water, ultraviolet absorbents in fibers, cosmetics, etc., after adsorption of titanium dioxide or zirconium oxide, etc.

[0041]

(4) The material of this invention is soluble in an organic solvent enabling it to be dispersed uniformly in plastic films, sheets, tubes, etc., providing plastics with a new function. /5

[0042]

For example, if the material of this invention is applied to a synthetic fiber for fabrics, it provides the fiber with a skin touch feel which is less sticky than the fiber prepared from the plastics alone, similar to natural fibers, and excellent performance with balanced moisture absorption and release as well as good heat retention and minimal dew formation is exhibited.

[0043]

Furthermore, porous rubbers such as latex vulcanized for solidification or urethane foam have been used to prepare a cosmetic puff, but those previous products have been unsatisfactory with respect to moist feel on the surface, skin touch feel, etc. If the material of this invention dissolved in an organic solvent is added at the time of urethane foam preparation, it is possible to prepare a sponge (puff) having a moist feel and smooth skin touch feel similar to those of silk. Incidentally, the production of urethane foam is carried out by using the same procedures as those used previously, and the amount of the material of this invention to be added is suitably selected depending on the purpose of application, etc., of the sponge to be prepared.

[0044]

(5) The material of this invention shows a performance attributable to functional groups such as carboxyl groups, etc., in the molecule, and consequently, it is applicable to various application fields. For example, the material of this invention having surfactant effects is applicable as a surfactant, fiber additive, antistatic agent, dye fixing agent, mordanting aid, etc. Furthermore, those esters of the material of this invention (its carboxyl group) with ascorbic acid, pantothenic acid, etc., are usable as a cosmetic raw material. The material of this invention is also usable as a raw material for chelating agents for capturing metal ions.

[0045]

In addition, the material of this invention is applicable to various application fields such as ink-jet recording paper, artificial skin, artificial organs, artificial leather, fixer for analytical and physicochemical tests, coating material for agrichemicals and fertilizers, L-B film-forming biosensors, artificial functional membranes (such as artificial cell membrane, etc.), microcapsule material, drug delivery system, etc.

[0046]

Application examples

This invention is explained in more detail below by using application examples.

[0047]

Application Example 1

In warm water at 35°C, 800 g of frozen egg white was left for defrosting, and after filtration through a 32 mesh filter, it was diluted 2.5 times with tap water, and it was adjusted to pH 6.8 with a 2 M solution of citric acid. The egg white solution prepared was filtered through a

42 mesh filter and centrifuged (7000 rpm X 10 min) to remove any precipitates formed after pH adjustment. The centrifugation supernatant obtained was applied to electrodialysis (electrodialysis equipment: Model CS-O, Asahi Glass K.K., flow rate of 250 L/h and specified voltage: 14 V, electroconductivity of 950 μS/cm). To the dialyzed solution, 1 N and 6 N aqueous solutions of sodium hydroxide were added to adjust to pH 10. After heating in boiling water for 30 min, the solution was cooled to a room temperature and filtered through a 100 mesh filter to obtain an egg white solution. The absorbance of the egg white solution prepared was 0.37 (280 nm), and the egg white concentration was about 4%.

[0048]

In 800 mL of the prepared egg white solution, 3 g of gelatin (Nitta Gelaltin K.K., molecular weight of 15,000) was dissolved, the solution was made up to 1 L, and 1 N and 6 N sodium hydroxide solutions were added to adjust to pH 12. The solution mixture was subsequently heated to 45°C. On the other hand, 18.4 g of toluene diisocyanate was dissolved in 300 g of chloroform, and the solution was added to the above solution mixture while being heated. Subsequently, after stirring for 2 h, the mixture was allowed to stand at room temperature, allowing it to separate into aqueous and chloroform phases. The aqueous phase was collected by centrifugation (8000 rpm X 10 min).

[0049]

If this supernatant is adjusted to pH 4 with citric acid, the material of this invention was precipitated, and the mixture was centrifuged (10,000 rpm X 15 min). The material of this invention isolated was easily soluble in DMF, and if the solution was poured into water, protein precipitates were formed. The precipitates were freeze-dried to obtain a powder which was also soluble in DMF, and if the solution prepared was poured again into water, protein precipitates were formed again.

[0050]

Application Example 2

Similarly to Application Example 1, 5-20 wt% of gelatin was added to the egg white to prepare the material of this invention.

[0051]

To 4 g of the material of this invention prepared, ionic [sic; deionized] water was added to make up to 1 L. While stirring, 1 N and 6 N aqueous solution of sodium hydroxide were added to adjust to pH 7 to dissolve the material of this invention in water. To the solution prepared,

1 mL of a paprika coloring matter (emulsion of natural paprika, trade name: Paprika base 150 manufactured by Sanei Kagaku Kogyo K.K.) was added, mixed, and after adjusting to pH 4 with a 2 M aqueous solution of citric acid, the mixture was allowed to stand at a room temperature to obtain the precipitates of the material of this invention with the paprika coloring matter adsorbed. After allowing to stand at room temperature for 2 h, the supernatant of the solution was used to measure absorbance (470 nm) to determine the residual quantity of the paprika coloring matter. The results obtained are shown in Table 1.

[0052]

For comparison, the same procedures as those used in Application Example 1 except no gelatin were carried out to obtain a functional protein material (that is, the material disclosed in the Japanese Kokai Patent Application No. Hei 4[1992]-53466), and the same subsequent procedures as those described above were carried out to determine the residual quantity of the paprika coloring matter. The results obtained are also shown in Table 1.

[0053]

Table 1

① 対鶏卵白のゼラチン量 (%)	② 吸光度
0	0.52
5	0.31
10	0.26
20	0.14

Key: 1 Amount of gelatin on the amount of chicken egg white (%)
2 Absorbance

[0054]

As is apparent from the results shown in Table 1, the higher the amount of gelatin added, the lower the residual quantity of the paprika coloring matter (the lower the absorbance). Therefore, the protective colloidal property of gelatin is considered to be exhibited, and it is presumed that the higher the amount of gelatin added, the higher the capacity of adsorption. Specifically, according to the process of this invention, it is possible to obtain a protein material having a function of being soluble in an organic solvent while retaining the original function of gelatin.

[0055]

Application Example 3 /6

To 2.5 g of the material of this invention prepared in Application Example 1, ionic water was added to make up to 1 L, and 1 N and 6 N aqueous solutions of sodium hydroxide were used to adjust to pH 7 to dissolve the material of this invention in water. To the solution prepared, 1.5 g of titanium dioxide (trade name: TAF-100, particle size: 0.05 μm, manufactured by Fuji Titanium Kogyo K.K.) was added, supersonic and disperser treatments were carried out for 10 min each to disperse the material of this invention and titanium dioxide, and subsequently, the dispersion was adjusted to pH 4 to cause the material of this invention with titanium dioxide adsorbed to precipitate. The precipitates were collected by centrifugation (10,000 rpm X 20 min) to obtain the material of this invention with titanium dioxide adsorbed.

[0056]

Incidentally, the supersonic generator used was a Branson 2200 (Yamato Kagaku K.K.), and the disperser used was an Ultradisperser, Model LK-22 (Yamato Kagaku K.K.)

[0057]

The material of this invention with titanium dioxide adsorbed was dispersed in ionic water, and ultraviolet absorption in the range of 200-400 nm was determined. The results obtained are shown in Figure 1. As apparent from the results shown in Figure 1, the material of this invention with titanium dioxide adsorbed is found to show an ultraviolet absorption spectrum similar to that of titanium dioxide itself. Therefore, it is apparent that the material of this invention has titanium dioxide adsorbed.

[0058]

Application Example 4

Nylon 66 chips were dissolved in formic acid by taking 1 day to obtain a 20% Nylon 66-formic acid solution (Solution A). Furthermore, the material of this invention prepared in Application Example 1 was also dissolved in formic acid to obtain a 3% functional protein-formic acid solution (Solution B).

[0059]

2 mL of the Solution B was added to 10 g of the Solution A, and the mixture was stirred to make a homogeneous mixture to obtain a solution of nylon containing about 3% of the material of this invention. The nylon solution prepared was spread on a flat glass plate and air-dried to disperse the solvent to obtain a white film having a weak tensile strength.

[0060]

The white film prepared was pressed and heated by using an impulse sealer (trade name: Impulse Sealer, Model F1-200-10w) to obtain a transparent nylon film having [a higher] tensile strength. The product prepared was found to possess a moisture-permeation property not found in a nylon only film.

[0061]

Application Example 5

In warm water at 35°C, 800 g of frozen egg white was left for 8 h for defrosting, after filtration through a 32 mesh filter, it was diluted 2.5 times with tap water, and it was adjusted to pH 6.8 with a 2 M solution of citric acid. The egg white solution prepared was filtered through a 42 mesh filter and centrifuged (7000 rpm X 10 min) to remove any precipitates formed after pH adjustment.

[0062]

The centrifugation supernatant obtained was applied to electrodialysis (electrodialysis equipment: Model CS-O, Asahi Glass K.K., flow rate of 250 L/h and specified voltage: 14 V, electroconductivity of 985 μS/cm). To the dialyzed solution, 1 N and 6 N aqueous solutions of sodium hydroxide were added to adjust to pH 10. After heating in boiling water for 30 min, the solution was cooled to room temperature and filtered through a 200 mesh filter to obtain an egg white solution having an absorbance of 0.386 (280 nm). The solution was diluted and adjusted to pH 12. The absorbance of the diluted egg white solution prepared was 0.163 (280 nm).

[0063]

In 400 mL of the diluted egg white solution prepared, 7 g of whey protein (trade name: DF-WPC, manufactured by Meiji Nyugyo K.K., partially skimmed whey protein) was dissolved, and the solution was adjusted again to pH 12. The absorbance in the case was 0.328 (280 nm). The mixture was heated to 45°C, 7 g of toluene diisocyanate and 300 g of chloroform were added, and the mixture was stirred for 2 h. After standing at a room temperature to allow the mixture to separate to aqueous and chloroform phases, the aqueous phase was collected. The aqueous phase collected was adjusted to pH 4 by adding citric acid to allow the material of this invention to precipitate, and the precipitates were collected by centrifugation (10,000 rpm X 30 min) to obtain the material of this invention.

[0064]

The material of this invention isolated was easily soluble in DMF, and if the solution was poured into water, protein precipitates were formed. The precipitates were freeze-dried to obtain a powder which was also soluble in DMF, and if the solution prepared was poured again into water, protein precipitates were formed again.

[0065]

Application Example 6

The solubility in a solvent (DMF) was examined for the material of this invention prepared from chicken egg white and gelatin in Application Example 1, material of this invention prepared from chicken egg white and whey protein in Application Example 5 and material prepared from chicken egg white alone (Application Example 2). The results obtained are shown in Table 2.

[0066]

Table 2

(1)素材の原料	素材濃度	溶解性(6)	
(2)鶏卵白+ホエー蛋白質	(5)40%	溶解後2日で白濁ゲル化	(7)
(3)鶏卵白+ゼラチン	20%	溶解後2日で白濁ゲル化	(8)
(4)鶏卵白	20%	溶解後2時間で白濁ゲル化	(9)

Key:
1 Raw material
2 Chicken egg white + whey protein
3 Chicken egg white + gelatin
4 Chicken egg white
5 Material concentration
6 Solubility
7 Becoming opaque and gelated in 2 days after dissolution
8 Becoming opaque and gelated in 2 days after dissolution
9 Becoming opaque and gelated in 2 h after dissolution

[0067]

As apparent from the results shown in Table 2, the concomitant use of two kinds of proteins allows the solubility of the protein material prepared in organic solvent to increase.

[0068]

Application Example 7

Ionic water was added to 4 g of the material of this invention prepared in Application Example 5 to make up to 1 L, and 1 N and 6 N aqueous solutions of sodium hydroxide were added while stirring to dissolve the material of this invention. To the solution prepared, 1 mL of Paprika coloring matter (Paprika Base 150) was added, mixed, adjusted to pH 3.3 by adding a 2 M citric acid solution, and the mixture was allowed to stand at room temperature to obtain precipitates of the material of this invention with the paprika coloring matter adsorbed. /7

[0069]

Application Example 8

In warm water at 35°C, 800 g of frozen chicken egg white was placed to thaw, the thawed egg white was filtered through a 32 mesh filter, diluted 4 times with tap water and centrifuged (7000 rpm X 10 min), and it was adjusted to pH 12. To 40 mL of the diluted egg white solution prepared, 7 g of whey protein (trade name: DF-WPC, manufactured by Meiji Nyugyo K.K., partially skimmed whey protein) was dissolved, and the solution was adjusted again to pH 12. The absorbance in the case was 0.355 (280 nm).

[0070]

The mixture was heated to 45°C, 7 g of toluene diisocyanate and 300 g of chloroform were added, and the mixture was stirred for 2 h. After standing at a room temperature to allow the mixture to separate into aqueous and chloroform phases, the aqueous phase was collected and centrifuged (8000 rpm X 10 min) to remove any impurities. The aqueous phase collected was adjusted to pH 3.5 by adding citric acid to allow the material of this invention to precipitate, and the precipitates were collected by centrifugation (8000 rpm X 10 min) to obtain the material of this invention. The material of this invention was found to be easily soluble in DMF and insoluble in water.

[0071]

Application Example 9

The same procedures as those used in Application Example 8 except that the mixture of 7 g of toluene diisocyanate and 300 g of chloroform in Application Example 8 was substituted with 10 g of toluene diisocyanate alone to obtain the material of this invention easily soluble in DMF but insoluble in water.

Brief description of the figure

Figure [1] is a drawing showing an ultraviolet absorption spectrum of an aggregate of the material of this invention and titanium dioxide in the range of 200-400 nm.

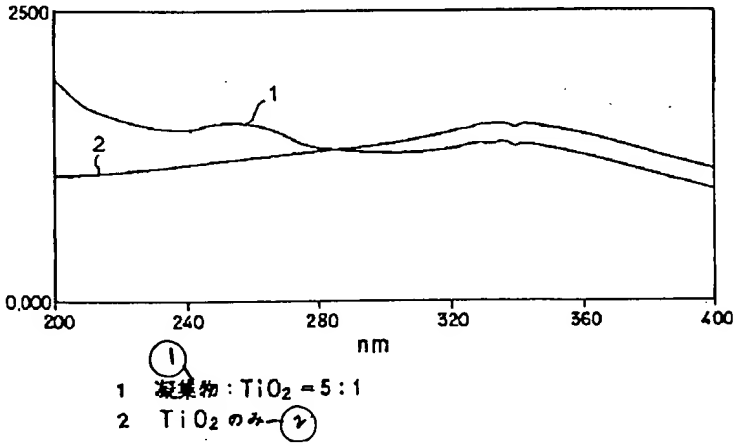

Figure 1. Ultraviolet absorption scanning in the range of 200-400 nm

1 Aggregate:

2 TiO_2 alone